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GENOMIC DNA METHYLATION PATTERNS AND SURVIVAL OF APPLE BUDS TO CRYOPRESERVATION

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Cryopreservation using the winter vegetative bud method (1) is being implemented in Asturias (Spain), a region with a mild oceanic climate. Two apple cultivars were assayed. Twigs with buds were collected from the field in February and March. Cultivar „De la Riega” taken on February gave high survival after cryopreservation but very low survival was obtained with the cultivar Raxao and no survival was achieved in samples of both cultivars when collected on March. So although it is feasible to apply this method to apples growing in mild temperatures, results differ between cultivars and the dormant bud stage seems to have a capital importance.

Experiments made with chestnut buds have shown a relationship between DNA cytosine methylation and bud dormancy (2). Hypermethylated DNA was found in dormant buds coinciding with unfavourable conditions for active growth, and DNA became increasingly hypomethylated during bud burst, when conditions are favourable for growth and development. Opposite patterns were found with the levels of acetylated histone H4 which were higher during bud burst than during bud dormancy. A similar work on global DNA methylation has been carried out in samples of these two apple cultivars in order to find out a correlation between responses of cultivars concerning survival after cryopreservation and the collection time. DNA was extracted from lyophilised buds excised from only desiccated scion woods (approx. 30% moisture content) and from cryopreserved scion woods and global methylation is quantified by High Performance Capillary Electrophoresis (3). The determination of global methylation from isolated DNAs takes less than 10 minutes per sample, so it can be a good tool for screening in case there is a correlation between methylation level and survival percentage to cryopreservation. Experiments need to be repeated as controversial results have been obtained.

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